

RATE MEASUREMENTS OF BIOMOLECULAR
REACTIONS USING ELECTROCHEMILUMINESCENCE

ABSTRACT

The rate of a biomolecular reaction, such as an enzymatic reaction or an affinity binding reaction, is measured using electrochemiluminescence ("ECL"). The reaction is conducted in an electrochemical cell with a mixture of reagents including a luminophore which will relate the concentration of a reactant, a reaction partner or the reaction product of a reaction partner to the ECL intensity. The reaction partner is a reagent which reacts with the reactant and which participates with the luminophore (or its reaction product participates with the luminophore) to cause the emission of ECL. The ECL intensity is modulated with a series of electrical pulses which are applied to the mixture of reagents at a preselected potential and for preselected intervals of time and duration. The ECL intensity is measured at the same intervals to provide a timed series of values (P). The same experiment is repeated except that the modulation is conducted after the reaction has gone to completion to obtain a timed series of values (C). The same experiment is repeated a third time in the absence of the reaction partner to obtain a times series of values (B). The results are normalized (N) using the following formula:

(I)
$$N = \frac{P-B}{C-B}$$

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to obtain a series of values N which can be used to plot the time course (concentration vs. time) of the reaction.

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